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      ANSWER 1 OF 26 CAPLUS COPYRIGHT 2003 ACS
      Multiplex PCR-microarray hybridization method for simultaneous
      detection of nucleic acid molecules
 SO
      PCT Int. Appl., 39 pp.
      CODEN: PIXXD2
      Schmidt, Wolfgang; Mundlein, Axel; Huber, Martin; Kroath, Hans
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     ANSWER 2 OF 26 CAPLUS COPYRIGHT 2003 ACS
     Detection of EHEC by hybridization or PCR analysis of Slt and eae and/or
 TΤ
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     PCT Int. Appl., 86 pp.
     CODEN: PIXXD2
     Grabowski, Reiner; Groenewald, Cordt; Schneider, Astrid; Pardigol,
 ΙN
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L3
     ANSWER 3 OF 26 CAPLUS COPYRIGHT 2003 ACS
TI
     Estimation of genetic divergence among elite cotton cultivars-genotypes
by
     DNA fingerprinting technology
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     Qin, Xuan; Turgeon, David K.; Ingersoll, Brian P.; Monsaas, Peter W.;
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     Lemoine, Christina J.; Tsosie, Treva; Stapp, Lynn O.; Abe, Patrick M.
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     A novel multiplex RT-PCR probe capture hybridization
TΙ
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- SO Journal of Virological Methods (1991), 35(2), 143-57 CODEN: JVMEDH; ISSN: 0166-0934
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  J.
- => d 13 1, 10, 12, abs
- L3 ANSWER 1 OF 26 CAPLUS COPYRIGHT 2003 ACS
- AB A method is disclosed for the simultaneous detection of at least two mutually different nucleic acid mols. in a sample. In a first step a multiplex PCR and in a second step a hybridization reaction are carried out on immobilized probes in a microarray. The hybridized PCR products are then detected and optionally quantified, whereby the probes applied for the hybridization reaction, which each hybridize specifically with nucleic acids which are different from each other, have m.ps. which differ from each other by at most 2 .degree.C, and preferably 1 .degree.C.

Thus, using Gene Runner software, probes with Tm of 65.+-.1.degree. were designed for PBP2, KanR, MecR, DhfrA, StrR, VanB, MlsR, AmpR, CmR, TetR, FosB, and AacA genes. Microarrays contg. these probes were then used to detect the microbial antibiotic resistance genes.

- L3 ANSWER 10 OF 26 CAPLUS COPYRIGHT 2003 ACS
- AB Rapid detection and characterization of food borne pathogens such as Escherichia coli O157:H7 is crucial for epidemiol. investigations and food

safety surveillance. As an alternative to conventional technologies, we examd. the sensitivity and specificity of nucleic acid microarrays for detecting and genotyping E. coli O157:H7. The array was composed of oligonucleotide probes (25-30 mer) complementary to four virulence loci (intimin, Shiga-like toxins I and II, and hemolysin A). Target DNA was amplified from whole cells or from purified DNA via single or multiplexed polymerase chain reaction (PCR), and PCR products were hybridized to the array without further modification or purifn. The array was 32-fold more sensitive than gel electrophoresis and capable of detecting amplification products from <1 cell equiv. of genomic DNA (1 fg). Immunomagnetic capture, PCR and a microarray were subsequently used to detect 55 CFU

ml-1

- (E. coli 0157:H7) from chicken rinsate without the aid of pre-enrichment. Four isolates of E. coli 0157:H7 and one isolate of 091:H2, for which genotypic data were available, were unambiguously genotyped with this array. Glass-based microarrays are relatively simple to construct and provide a rapid and sensitive means to detect multiplexed PCR products; the system is amenable to automation.
- L3 ANSWER 12 OF 26 CAPLUS COPYRIGHT 2003 ACS
  AB A PCR-based method for detecting the presence of multiple virus infections

in a biol. sample, PCR-enzyme hybridization assay, or PCR-EHA is disclosed. In one embodiment, this method comprises the step of (1) isolating a nucleic acid either in the form of RNA or cDNA derived from

it, (2) exposing the nucleic acid to a primer pair derived from the sequences of human parainfluenza virus 1, 2 and 3, respiratory syncytial virus A and B and influenza virus, A and B to carry out a PCR amplification, and (3) examg. the amplification reaction product using

protein-linked oligonucleotide probes attached to a sold support. In another embodiment, the invention is an improved PCR method. where the 5' and 3' primers of unequal concns. are used to improve the detection.

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DOCUMENT NUMBER:

132:89219

TITLE:

the

Multiplex PCR-enzyme hybridization assay

using unequal primer concentrations to detect human parainfluenza virus 1, 2, 3 and respiratory syncytial

virus A, B and influenza virus A, B

Henrickson, Kelly J.; Fan, Jiang

INVENTOR(S): PATENT ASSIGNEE(S):

MCW Research Foundation, USA

SOURCE:

U.S., 26 pp., Cont.-in-part of U.S. 5,744,299.

CODEN: USXXAM

DOCUMENT TYPE:

Patent English

LANGUAGE: FAMILY ACC. NUM. COUNT:

PATENT INFORMATION:

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US							US 1996-691045					10060001					
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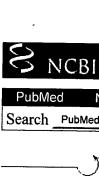
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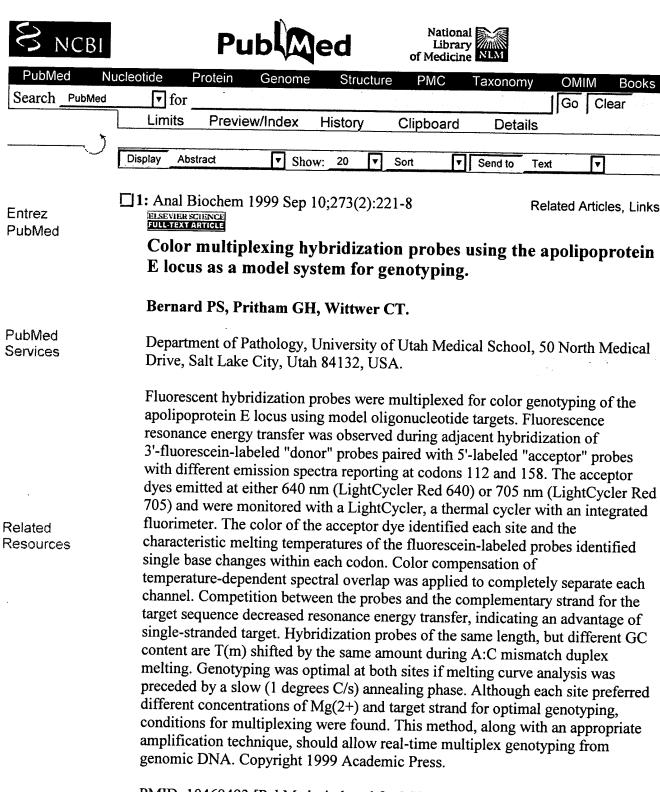
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DNA-EIA to detect high and low risk HPV genotypes in cervical lesions with E6/E7 primer mediated multiplex PCR.

Clavel C, Rihet S, Masure M, Chypre C, Boulanger JC, Quereux C, Birembaut P.

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BACKGROUND: Oncogenicity of human papillomavirus (HPV) DNA in

premalignant and malignant uterine cervical diseases is mainly induced by E6/E7 open reading frame (ORF). The presence of an oncogenic HPV DNA may be a

diagnostic marker for the detection of cytologically negative smears. AIMS: To

evaluate an original polymerase chain reaction enzyme immunoassay (PCR-EIA) for the detection and typing of oncogenic and non-oncogenic HPV types. METHODS: The test was an original multiplex labelled PCR-EIA for the detection and typing of oncogenic and non-oncogenic HPV using three consensus sequence primers within the oncogenic E6/E7 ORF. One primer was dinitrophenyl (DNP) labelled and the DNP labelled amplimers could be further hybridised with specific biotinylated oligoprobes mixed in only two cocktails: oncogenic (16, 18, 31, 33, 35, 52, and 58) and non-oncogenic (6 and 11) HPV types in only two wells; then biotinylated oligoprobes were deposited in streptavidin-coated microplates. The PCR-EIA was validated on HPV plasmids (types 6, 11, 16, 18, 31, 35, 52, and 58) and used to evaluate cervical scrapes from 181 patients (median age 32 years) at high risk for cervical cancer. RESULTS: HPV were detected in the cervical scrapes of 88 of 181 patients (48.6%); nine with non-oncogenic HPV (5.0%) and 79 with oncogenic HPV (43.6%) including 29 coinfections with oncogenic and non-oncogenic HPV. The number of oncogenic HPV infections increased with the presence of high grade lesions: 95.8% of the cervical scrapes from patients with high grade lesions contained oncogenic HPV compared with 32.1% of the specimens from patients without any lesions detectable by colposcopy and/or by cytological examination of the cervical smears. Moreover, 60% of cervical scrapes exhibiting low grade lesions contained oncogenic HPV. CONCLUSIONS: This test is simple, specific, sensitive, safe, fast, reproducible, and easy to use in routine practice. Thus, it is possible to detect simultaneously on a simple cervical scrape, two kinds of HPV--oncogenic and non-oncogenic--in just two microplate wells with non-isotopic oligoprobes.